

AMENDMENTS TO THE SPECIFICATION

Please replace the paragraph starting on page 5, line 19 of the specification as filed with the following rewritten paragraph:

A test agent which has an effect on expression of CYP2S1, may do so by binding to the nucleic acid responsible for controlling CYP2S1 expression. Typically, such a test agent binds to regulatory sequences found upstream of the CYP2S1 gene sequence and the present invention provides a 10 kilo base region of such a sequence, in which a number of putative regulatory domains are identified. The 10 kilo base sequence (SEQ ID NO:10) shown in Figure 7 can be used to affect translation of CYP2S1 or a suitable reporter protein known to those skilled in the art.[[.]] It is also relatively straightforward for the skilled man to produce deletion mutants of this sequence and test for their ability to still affect expression of CYP2S1 or a reporter protein.

Please replace the paragraph starting on page 10, line 5 of the specification as filed with the following rewritten paragraph:

Figure 7 shows 10kb of nucleotide sequence upstream from the ATG start site for CYP2S1 (SEQ ID NO:10).

Please replace the paragraph starting on page 11, line 19 of the specification as filed with the following rewritten paragraph:

Sequence-specific primers and probes for Taqman quantitative PCR analysis of CYP2S1 mRNA expression were designed using PE Applied Biosystems Primer Express software, according to the manufacturer's protocol. PCR (1 x (50°C, 2 minutes, 95°C, 10 minutes), 40 x (92°C, 15 seconds, 60°C, 1 minute)) was performed in the presence of 0.6 x Taqman Universal PCR Master Mix (PE Applied Biosystems), 300nM forward primer (5'-CGA TGC CTT CCT GCT GAA G-3' SEQ ID NO:1), 300nM reverse primer (5'-GCA TGT TCT TGT TGG TGA ATT CTG 3' SEQ ID NO:2) and 175 nM fluorescent probe (5'-FAM-TGG CAC AGG AGG AAC AAA ACC CAG G-3'

SEQ ID NO:3). The assay was designed such that the probe spanned an intron/exon boundary to minimise the possibility of co-amplifying genomic DNA. Similarly, assays were designed for CPR (Forward primer 5' CCT GCA GGC CCG CTA CTA 3' SEQ ID NO:4, Reverse primer 5' TTG GTC TCG TAC TCC ACA ACC A 3' SEQ ID NO:5, Probe 5'-FAM- TCC TCC AAG GTC CAC CCC AAC TCT GT 3' SEQ ID NO:6) and CYP26 (Forward primer 5' CCG TAT TTC CTG CGC TTC AT 3' SEQ ID NO:7, Reverse primer 5' TTC CCC TTC TTT GGG GAA AC 3' SEQ ID NO:8, Probe 5'-FAM- CAG GAA CTT CCT CCG CTG CAG TAC CAT 3' SEQ ID NO:9). Real-time PCR was performed on an ABI Prism 7700 Sequence Detector, where fluorescent output was directly proportional to input cDNA concentration. Input cDNA concentrations were normalised to 18S ribosomal RNA, using PE Applied Biosystems Ribosomal RNA control reagents. Oligonucleotide primers were synthesised by MWG Biotech and fluorescent Taqman probes by PE Applied Biosystems.

Please replace the paragraph starting on page 14, line 22 of the specification as filed with the following rewritten paragraph:

In order to investigate possible mechanisms of CYP2S1 regulation, the inventors examined sequences in the CYP2S1 gene promoter to identify regulatory elements which may influence CYP2S1 expression and inducibility. No CYP2S1 promoter sequence has been published to date, but the CYP2S1 gene has been mapped to chromosome 19q13.2¹³. The inventors therefore identified the CYP2S1 gene promoter by mapping the CYP2S1 cDNA onto a genomic clone (NCBI Accession number NT_011139) containing the CYP450 cluster on chromosome 19¹⁴. Recent evidence suggests that transcriptional regulation of CYP450 genes can involve distal regulatory sequences, with a potent enhancer sequence as far as 8kb distal to the transcription start¹⁸. In light of these findings, the inventors analysed a 10kb fragment of the CYP2S1 promoter (SEQ ID NO:10), including the sequence immediately preceding and including the transcription start site.